

CLAIMS

1. A binding motif capable of binding to a cytoplasmic protein said motif consisting of the following amino acid sequence:

5 . N-X-X-Y

wherein X is any residue, and Y is tyrosine or an equivalent thereof.

2. A binding motif of a receptor molecule capable of binding to a cytoplasmic protein said motif consisting of the following amino acid sequence:

10 N-X-X-Y

wherein X is any residue, and Y is tyrosine or an equivalent thereof.

3. A binding motif according to claim 1 or 2 derived from a receptor selected from the group including:

- | | | |
|----|------|---|
| 15 | (1) | GM-CSF/IL-3/IL-5 receptor |
| | (2) | IL6 human interleukin-6 receptor beta chain precursor (IL-6R-beta) |
| | (3) | LEPR human leptin receptor precursor (LEP-R) (OB RECEPTOR) (OB-R). |
| | (4) | TNR2 human tumor necrosis factor receptor 2 precursor (tumor necrosis |
| 20 | | factor |
| | (5) | VGR1 human vascular endothelial growth factor receptor 1 precursor |
| | (6) | TRK3 human receptor protein-tyrosine kinase TKT precursor (EC 2.7.1.112) |
| | (7) | Q01974 protein-tyrosine kinase transmembrane receptor ROR2 |
| 25 | | precursor |
| | (8) | FGR1 human basic fibroblast growth factor receptor 1 precursor (BFGF-R) |
| | (9) | Q15426 protein-tyrosine phosphatase, receptor-type, H precursor (EC 3.1.3.48) |
| 30 | (10) | PTPM human protein-tyrosine phosphatase mu precursor (EC 3.1.3.48) (R-PTP-MU). |
| | (11) | PDGS human alpha platelet-derived growth factor receptor precursor (EC 2.7.1.112) |

- (12) FGR4 human fibroblast growth factor receptor 4 precursor (FGFR-4) (EC 2.7.1.112)
- 5 (13) FGR2 human fibroblast growth factor receptor 2 precursor (FGFR-2) (EC 2.7.1.112)
- (14) Q13635 patched protein homolog (PTC)
- (15) MANR human macrophage mannose receptor precursor.
- (16) LRP2 human low-density lipoprotein receptor-related protein 2 precursor (megalin)
- 10 (17) IDD human integral membrane protein dgcr2/idd precursor (KIAA0163)
- (18) AMFR human autocrine motility factor receptor precursor (AMF receptor) (gp78)
- (19) ACH5 human neuronal acetylcholine receptor protein, alpha-5 chain precursor.
- 15 (20) KKIT human: stem cell growth factor receptor (proto-oncogene tyrosine-protein kinase kit) (C-KIT) (CD117 antigen)
- (21) TPOR human: thrombopoietin receptor precursor (TPO-R) (myeloproliferative leukemia protein (C-MPL). TPOR or MPL.
- (22) TPOR mouse: thrombopoietin receptor precursor (TPO-R)
- 20 (myeloproliferative leukemia protein) (C-MPL). TPOR or MPL.
- (23) Acetylcholine R
- (24) Acetylcholine R alpha-5
- (25) C-C chemokine receptor 6
- (26) Middle T antigen
- 25 (27) integrin alpha 1
- (28) FGFR2 (KGF R)
- (29) FGFR1 (flg)
- (30) FGFR5
- (31) Erb4
- 30 (32) Vaccinia virus protein A36R
- (33) Macrophage mannose R (MRC1)
- (34) LDLR
- (35) VLDL (rat)

- (36) LRP1 low density lipoprotein receptor-related protein 1
 - (37) integrin beta 1
 - (38) integrin beta 7
 - (39) integrin beta 3
 - 5 (40) integrin beta 5
 - (41) integrin beta 6
 - (42) G-CSFR1 (second)
 - (43) G-CSFR1
 - (44) g-csf-r
 - 10 (45) IL-6B (gp130)
 - (46) LeptinR
 - (47) ProlactinR
 - (48) insulinR
 - (49) irs-1
 - 15 (50) IGFI R
 - (51) flt3 R
 - (52) VEGFR2 (FLK1)
 - (53) PDGF R-alpha
 - (54) IL-9R
 - 20 (55) BetaR
- or a functional equivalent or analogue thereof.

4. A binding motif or equivalent thereof according to any one of claims 1 to 3 derived from a sequence selected from the group including:

- 25 NGPYLG.....PP..HSRSLP
- NVHYRT.....P...KTHTMP
- **RYFTQKEE.....TESGSGP
- NKKYELQDRDVCE....P.RYRSVSEP
- NPTY SVM.....RSHSYP
- 30 NIFYLIR...KSGSFPMP ELKLSISFP
- NEEYLDLSQ.....PLEQYSPSYP
- NQEYLDLSM.....PLDQYSPSFP
- NATYKVD.....VIQRTRSKP
- NPEY.....HSASSGP
- 35 NPDY.....WNHSLP
- NPSYSSNP FVNYN....KTSICKSNP
- NTLY.....FNSQSSP
- NPVYQKTTEDEVHI...CHNQDGYSYP

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NPVYLKTTEEDLSIDIG..RH.SASVG
 NPTYKMYEGGEPDDVGGLLDADFALDPDKPTNFTNPVY
 NPIY.....KSAVTTTV
 NPLY.....KSAITTTV
 5 NPLY.....KEATSTFT
 NPLY.....RKPISTHT
 NPLY.....RGSTSTFK
 PGHYL.....RCDSTQP
 VQTYVLQ.....GDPRAVSTQP
 10 QVLYGQLL.....GSPTSP
 HSGYRHQVPSVQVF.....SRSESTQP
 WKMYEVYDA.....KS.KSVSLP
 KIPYFHA.....GGS.KCSTWP
 ELDYCLKGLKL.....P.S.RTWSPP
 15 SGDYMPPM.....SPKSVSAP
 SFYYSEENKLPEPEELDLEPENMESVP(LDPSASSSSSLP)
 EEIYIIM.....QSCWAFDSRKRPSPF
 ISQYLQN.....S.KRKSRP
 GTAY.....GLSRSQP
 20 ***YLPQEDWAP.....TSLTRP
 LVAYIAFKRWNSCKQN...KQGANSRPVNQTTPPEGEKLHSDSGIS

5. A binding motif according to any one of claims 1 to 4 having a sequence selected from the group including:

25 NGPY
 NVHY
 **RY
 NKKY
 NPTY
 30 NIFY
 NEEY
 NQEY
 NATY
 NPEY
 35 NPDY
 NPSY
 NTLY
 NPVY
 NPIY
 40 NPLY

6. A binding motif according to any one of claims 1 to 5 wherein the sequence is derived from a cytokine receptor.

45 7. A binding motif according to any one of claims 1 to 6 wherein the receptor is the GM-CSF/IL-3/IL-5 receptor.

8. A binding motif according to any one of claims 1 to 7 wherein the sequence includes the common beta chain (β c).

5 9. A binding motif according to any one of claims 1 to 8 wherein the Tyr residue is equivalent to Tyr577 of the common beta chain (β c).

10. A binding motif according to any one of claims 1 to 9 having a modification at a residue equivalent to the Tyr residue.

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11. A binding motif according to any one of claims 1 to 10 wherein the residue equivalent to the Tyr residue is substituted with a Phe residue.

12. A binding motif according to any one of claims 1 to 11 which binds to a
15 cytoplasmic protein selected from the group including 14-3-3 protein, Shc, SHIP-2, WW-domain of the prolyl isomerase, Pin1 and the ubiquitin ligase, NEDD4.

13. A binding motif according to any one of claims 1 to 12 wherein the
20 cytoplasmic protein is Shc or SHIP-2.

14. A method of modulating activity in a cell, said method including:
introducing a modification to a binding motif capable of binding to a
cytoplasmic protein said motif consisting of the following amino acid sequence:

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N-X-X-Y

wherein X is any residue, and Y is tyrosine.

15. A method of modulating activity in a cell, said method including:
introducing a modification to a binding motif according to any one of
30 claims 1 to 13.

16. A method according to claim 15 wherein the tyrosine residue is equivalent to Tyr577 of the common beta chain (β c).

17. A method according to claim 16 wherein the common beta chain (β c) is of the GM-CSF/IL-3/IL-5 receptor.

5 18. A method according to any one of claims 15 to 17 wherein the activity is modulated by introducing a modification of phosphorylation of the Tyr of the motif.

10 19. A method according to claim 18 wherein the phosphorylation is increased by subjecting the cell to a phosphorylating agent.

20. A method according to claim 19 wherein the phosphorylating agent is a kinase.

15 21. A method according to claim 18 wherein the phosphorylation is decreased by mutating, substituting or deleting the Tyr.

22. A method according to claim 23 wherein the Tyr is substituted for Phe.

20 23. A method according to claim 18 wherein the phosphorylation is decreased by subjecting the cell to an antagonist which inhibits phosphorylation of the Tyr.

25 24. A method according to claim 18 wherein the phosphorylation is decreased by subjecting the cell to a kinase inhibitor to inhibit phosphorylation of the Tyr.

30 25. A method according to any one of claims 21 to 24 for inhibiting cellular activity, said method comprising decreasing or inhibiting phosphorylation of the Tyr motif.

26. A method according to claim 25 for inhibiting activity in a cell, said method comprising inhibiting binding of a cytoplasmic protein to the motif.

27. A method according to claim 26 wherein the cytoplasmic protein is selected from the group including 14-3-3 protein, Shc, SHIP-2, WW-domain of the prolyl isomerase, Pin1 and the ubiquitin ligase, NEDD4.

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28. A method according to claim 27 wherein the cytoplasmic protein is Shc.

29. A method according to claim 19 or 20 for activating cellular activity, said method comprising inducing phosphorylation of the Tyr of the motif.

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30. A method according to any one of claims 14 to 29 wherein the cellular activity is selected from the group including cell survival, proliferation, differentiation, mitogenesis, transformation, chemotaxis, motility, enhanced phagocytosis, enhanced bacterial killing, superoxide production and cytotoxicity.

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31. A method according to any one of claims 14 to 30 wherein the cellular activity is cell survival.

32. A method according to any one of claims 14 to 30 wherein the cellular activity is proliferation.

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33. A method according to any one of claims 14 to 32 wherein the cell is a haematopoietic cell.

34. A method according to claim 32 for increasing proliferation, said method including inhibiting phosphorylation of the Tyr.

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35. A method according to claim 32 for inhibiting proliferation, said method including inducing phosphorylation of the Tyr.

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36. A method according to claim 32 for increasing cell growth, said method including inhibiting activation of the Tyr.

37. A method of modulating activity in a cell, said method including:
introducing a modification to a binding motif of a receptor capable of
binding to a cytoplasmic protein said motif consisting of the following amino acid
5 sequence:

N-X-X-Y

wherein X is any residue, and Y is tyrosine.

38. A method according to claim 34 wherein the tyrosine residue is
10 equivalent to Tyr577 of the common beta chain (β c).

39. A method according to claim 35 wherein the common beta chain (β c) is
of the GM-CSF/IL-3/IL-5 receptor.

15 40. A method according to claim 37 for increasing proliferation, said method
including inhibiting phosphorylation of the Tyr.

41. A method according to claim 37 for inhibiting proliferation, said method
including inducing phosphorylation of the Tyr.

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42. A method according to claim 37 for increasing cell growth, said method
including inhibiting activation of the Tyr.

25 43. A method for transplantation of cells, said method including
inhibiting activation of a Tyr of a binding motif according to any one of
claims 1 to 13 in the cells; and
transplanting the cells.

30 44. A method for enhancing transplantation efficiency, said method including
inhibiting activation of a Tyr of a binding motif according to any one of
claims 1 to 13 in the cells; and
transplanting the cells.

45. A method according to claim 43 or 44 wherein activation of the Tyr is inhibited *in vitro* prior to transplanting.

5 46. A method according to claim 43 or 44 wherein the activation of the Tyr is inhibited in a region of transplantation.

47. A method according to any one of claims 43 to 46 wherein activation is inhibited by inhibiting phosphorylation of the Tyr.

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48. A method according to any one of claims 43 to 47 wherein the Tyr is equivalent to Tyr577 of the common beta chain (β c) of the GM-CSF/IL-3/IL-5 receptor.

15 49. A method according to claim 48 wherein the common beta chain (β c) is of the GM-CSF/IL-3/IL-5 receptor.

50. A method of improving wound healing in a patient, said method including inhibiting activation of a Tyr of a binding motif according to any one of
20 claims 1 to 13 in a region of the wound.

51. A method according to claim 36 wherein the activation is inhibited by inhibiting phosphorylation of the Tyr.

25 52. A method according to claim 50 or 51 wherein the Tyr is equivalent to Tyr577 of the common beta chain (β c) of the GM-CSF/IL-3/IL-5 receptor.

53. A method according to claim 52 wherein the common beta chain (β c) is of the GM-CSF/IL-3/IL-5 receptor.

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54. A use of an inhibitor of activation of a Tyr of a binding motif according to any one of claims 1 to 13 in the preparation of a medicament for the treatment of a wound.

55. A use according to claim 54 wherein the inhibitor inhibits phosphorylation of the Tyr.

5 56. A use according to claim 54 or 55 wherein the Tyr is equivalent to Tyr577 of the common beta chain (β c).

57. A use according to claim 56 wherein the common beta chain (β c) is of the GM-CSF/IL-3/IL-5 receptor.

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58. A method for screening of cell growth promoting compounds, said method including

obtaining a cell having a receptor containing a β c having a Tyr577 residue or equivalent;

15 inducing phosphorylation of the Tyr or an equivalent in a binding motif according to any one of claims 1 to 13;

exposing the cell to the compound; and

assessing colony formation of the cells.

20 59. A method according to claim 58 wherein the Tyr is equivalent to Tyr577 of the common beta chain (β c).

60. A use according to claim 56 wherein the common beta chain (β c) is of the GM-CSF/IL-3/IL-5 receptor.